Wohl Virion Centre Annual Report 2012.

The Centre is located in the Cruciform building and comprises five PIs, eight postdoctoral fellows, one research assistant and four PhD students. This year Prof. Paul Kellam with his UCL group joined the Wohl, bringing new expertise in virus genome sequencing and host-pathogens interactions. Grant income comes from the MRC, EPSRC, EU, Bill & Melinda Gates Foundation, Wellcome Trust, Biomedical Research Centre (BRC). Lab meetings are held every Thursday and are attended by all the members of the Centre.

Five groups work in the Centre:

Robin A Weiss PhD FRS

As an emeritus professor, he still works at least three days per week in the Wohl Virion Centre and currently has a lab comprising two post-doctoral researchers: Laura McCoy and Ben Webb. During 2012 former lab member Carolyn Edwards moved to a position at a Biotechnology company in Cambridge. Robin's research is supported by grants won competitively from the Bill & Melinda Gates Foundation, the Medical Research Council, the Engineering & Physical Sciences Research Council, and the European Union Framework 7 Programme. The research is collaborative with the University of Utrecht, the London Centre for Nanotechnology, and the University of KwaZulu-Natal.

Robin's interest focuses on the properties of the envelope of the AIDS virus, HIV, and on how we can apply our discoveries to the development of vaccines and diagnostic reagents affordable in resource-poor countries. The group has developed novel approaches to the neutralisation of HIV by antibodies and also on how HIV interacts with cell surface receptors. In particular, he exploited a curiosity of nature, namely that llamas make single chain antibodies, which we exploit to characterise the 'Achilles heels' of the HIV envelope. The expertise that we have developed on HIV has allowed us to apply similar techniques to the neutralisation of other enveloped viruses such as SARS, H5N1 influenza and rabies. The incorporation of a high containment laboratory for dangerous viruses in the Wohl Virion Centre made these investigations possible.

During the past year, Dr Laura McCoy represented the Wohl Virion Centre at the Keystone HIV Vaccines meeting in Keystone, USA, and the International AIDS Vaccine Conference in Boston, USA. She was also invited to speak at a British Society of Immunology Cambridge regional group seminar, hosted by the Department of Pathology at the University of Cambridge. Laura's paper describing a llama antibody which neutralises 96% of HIV strains tested was published in the June issue of The Journal of Experimental Medicine. A review article entitled "Neutralizing antibodies to HIV-1 induced by immunization" written by Laura and Robin will be published shortly in the same journal. In addition, both are co-authors on a manuscript describing a llama antibody targetting the HIV gp41 protein that will be published PLOS Pathogens. In August Laura received a Bill & Melinda Gates Foundation Young Investigator Award and an MRC Centenary Award Travel Fellowship via the UCL/MRC Centre for Medical Molecular Virology.

Clare Jolly PhD

Clare Jolly is an MRC Career Development Fellow who joined the Wohl Virion Centre in 2009. Her lab comprises two postdoctoral fellows, Dr Elisabetta Groppelli and Dr Alice Len and a MRC funded PhD student, Shimona Starling. Her work is funded by a MRC Career Development Award and a MRC project grant. The research interests of the Jolly laboratory are directed at understanding the molecular mechanisms that regulate Human Immunodeficiency Virus transmission between CD4+ T cells – the main targets for the virus *in vivo*. This mode of HIV-1 transmission is important because it escapes most antiretroviral drugs developed so far and may be one of the main mechanisms to allow virus spread in lymphoid organs and during reactivation from latency. In particular Clare's group is interested in how the virus hijacks and possibly modifies existing cellular machinery to promote efficient transmission by direct cell-to-cell spread. Such

host factors may represent novel targets for therapeutic development. She has recently shown that HIV infected T cells can respond to contact with an uninfected T cell by polarizing the cellular cytoskeleton and secretory organelles towards the contact site. This work was published in 2011 in the open access journal PloS Pathogens and additional publications are being prepared for submission. Dr Jolly has recently given invited seminars at the Institut Cochin in Paris, King's College London and Imperial College London. She was an invited speaker at the 2012 Keystone Symposia meeting on Frontiers in HIV Pathogenesis, Therapy and Eradication in Canada and co-organised the annual UK Recently Independent Virology Researchers (RIVR) in Derby 2013.

Yasuhiro Takeuchi PhD

Yasu Takeuchi's group comprises three PhD students, Sabine Winkler (mainly based at the NIBSC), Khaled Samber and Kanayo Doi (started in June 2012, awarded a highly-competitive full-studentship from Japan Student Services Organization). Yasu's main interests are in gene therapy and in xenotransplantation.

Yasu's work in gene therapy is in collaboration with Mary Collins and their most recent contribution was published in J. Virol (Knight et al 2012): the work addresses the problem of insertional mutagenesis caused by retroviral vectors by introducing a silencing-resistant promoter (UCOE) in self-inactivating lentiviral vectors (LV), which reduces their mutagenic activity. A related project, the development of stable LV packaging cell lines for future clinical application, has resulted in a patent being filed (Inventors: Sean Knight, Yasu Takeuchi and Mary Collins).

Takeuchi is a leader in the field of porcine endogenous retroviruses (PERV) and xenotransplantation and he studies the risks associated with potential transmission of such viruses to humans following transplantation of pig organs. Various studies on safety in xenotransplantation funded by EU FP6 XENOME consortium in which Takeuchi acted as the work package leader on safety (funding duration ended in April 2012) are now being published. Takeuchi was invited to the WHO consultation on xenotransplantation safety in 2011 where he delivered a paper entitled "Xenotransplantation-associated Infectious Risk: Considerations; Xenotransplantation" jointly with Linda Scobie from Glasgow and Jay Fishman from Boston. This paper was published in 2012. This was followed by two other publications: first a Nature article on the pig genome sequence in which Yasu, as one of 16 leading authors, was in charge of the PERV section; second a hepatitis E virus study published in Emerging Infectious Diseases. Further major publications are expected in 2013, including studies on burn patients who had received vital pigskin transplantation and safety studies in pig-to-primate xenotransplantation trials.

Ariberto Fassati MD PhD

Ari Fassati is the Director of the Wohl Virion Centre. His group comprises an MRC funded postdoctoral fellow, Alex Zhyvoloup, an EU FP7 Innovative Medicine-funded postdoctoral fellow, Aksana Labokha, one undergraduate student, Jun Low, and Nan-Yu Chen, a paediatrician with expertise in infectious diseases who won a Governmental scholarship from Taiwan and joined the Wohl Virion Centre as a PhD student in 2011. Ari also co-supervises two PhD students mainly based at the London Centre for Nanotechnology: Dino Osmanovic and Aizhan Bestembayeva. He collaborates with Bart Hoogenboom (London Centre for Nanotechnology), Stephan Beck (UCL Cancer Institute), Liz Murchison and Michael Stratton (Wellcome Trust Sanger Institute) and David Selwood, Medicinal Chemistry at UCL.

The Fassati lab has three main interests: the discovery of new antiviral drug targets, understanding the mechanisms of viral nuclear transport, and understanding the mechanisms of regression of the canine transmissible venereal tumour (CTVT) as a model to understand cancer regression in general.

In 2010 Fassati, with MRC funding, set up a high through put cell based screening facility in the Wohl Virion Centre to perform screenings with live pathogens. In 2012 the first two screening

projects were completed and the group made several discoveries; First, Hsp90 is required for HIV-1 gene expression and reactivation during hyperthermia (PLoS Path. 2012). Second, a transcription factor that induces histone acetylation important for HIV-1 gene expression has been identified by chemical genetics. A compound that inhibits the transcription factor has been identified (manuscript in preparation). Third, in collaboration with Robin Weiss and Laura McCoy, a high through put screening was performed and two new llama nanobodies with very broad and potent HIV-1 neutralizing activity have been isolated. Importantly, such nanobodies appear to target the CD4 receptor using the same "angle of approach" of the very broad human neutralizing antibodies recently identified (VCR01, VCR02). A patent has been filed on these new nanobodies.

On the nuclear import side, in collaboration with Benny Chain, Fassati showed that the nuclear import receptor importin 7, already implicated in nuclear transport of HIV-1, is also critical for nuclear import of transfected DNA. Furthermore, Chain and Fassati showed that importin 7 is part of an endogenous pathway for in mammalian cells for efficient accumulation of exogenous and endogenous DNA in the nucleus, which may be critical for the exchange of genetic information between mitochondria and nuclear genomes and to control activation of the innate immune response (Traffic 2012).

In collaboration with Bart Hoogenboom, Fassati is elucidating the biophysical nature of the selective barrier present in nuclear pores using biochemical methods, atomic force microscopy and mathematical modelling. The first paper describing the model was recently published (Phys Rev E 2012).

In collaboration with Stephan Beck, Liz Murchison and Mike Stratton, the Fassati lab has identified approximately 300 genes that are differentially expressed in progressive or regressive CTVT and mapped epigenetic changes of such genes, which will help to understand the mechanisms leading to its regression (ongoing work).

Ari is co-organising the 2013 meeting “Frontiers of Retrovirology” in Cambridge. He was invited speaker at the LMB Cambridge, and Brunel University. With Helen Weiss at the LSHTM, he is leading the HIV/AIDS research strategy of the new Bloomsbury Research Institute.

Paul Kellam PhD

Paul Kellam is the Viral Genomics group leader and Senior Investigator at the Wellcome Trust Sanger Institute and a Professor of Viral Pathogenesis in the Wohl Virion Centre. Paul was one of the original members of the Wohl Virion Centre when it was established in 1999. Paul’s work is funded by the Wellcome Trust, EU, MRC and BBSRC. Paul’s research focuses on the genetics and genomics of host and virus interactions. The laboratory is one of a small number of virology groups that combines molecular biology and virology with computational research to address basic biological questions in infection and immunity (Archer et al). Paul’s team in the Wohl Virion Centre consists of Dr Eve Coulter, Dr Ed Tsao and Dr Dan Frampton. At UCL Paul’s group investigates the B cell biology of Kaposi’s sarcoma associated herpesvirus (KSHV), a virus that is still the leading cause of cancer in Africa in HIV infected populations. In the Wohl Virion Centre Paul’s laboratory developed the first KSHV gene expression microarray to explore virus lytic replication and pioneered the use of host gene expression arrays to characterise herpesvirus driven B-cell tumours. This identified the B-cell differentiation transcription factor, X-box binding protein-1 (XBP-1) as the host transcription factor that switches KSHV from latency to the virus lytic cycle. This work continues now, exploring how B cell transcription factors control the KSHV life cycle (Coulter manuscript in preparation).

In 2009 Paul established the Virus Genomics laboratory at the Wellcome Trust Sanger Institute whilst maintaining his lab at UCL. The work of the Sanger Institute lab uses second generation sequencing technology to detect genetic variation of host and virus and uses this

knowledge to determine the molecular and pathogenic outcomes of virus infections (Wash et al). This work focuses on viruses of medical importance such as HIV (Gall et al) and influenza (Reperant et al; Ballie et al, Murica et al, Hughes et al and Lycett et al). Recently, we identified the first human influenza disease severity determining allele in the gene IFITM3 in people hospitalised with pandemic influenza A H1N1 (Everitt et al). In partnership with UCL and the Wohl Virion centre we are now starting a major new initiative to use second generation sequencing technology in clincal virology as part of the UK initiative on the application of genetics to patients with infectious disease.

Publications in 2012 from members of the Wohl Virion Centre (Centre members in bold).

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